Behavior and peripheral amine concentrations in relation to ractopamine feeding, sex, and social rank of finishing pigs[1]

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ABSTRACT: Aggression can impair productivity and well-being. The association between aggression in finishing pigs and the feed additive ractopamine (RAC), a β-adrenoreceptor agonist, is unknown and warrants further investigation. Our goal was to examine behavioral activity, including aggression, in the home pen and concentrations of peripheral amines in barrows and gilts, taking into account diet (RAC) and social rank. Sixty-four finishing pigs, housed in pens of 4 by sex, were fed either a control (CTL) or RAC-added (5 mg/kg for 2 wk plus 10 mg/kg for another 2 wk) diet. The top dominant and bottom subordinate pigs in each pen were determined at mixing (2 wk pretrial). The behavior of all pigs was recorded continuously during the pretrial week (baseline) and for the following 4 wk. These behavioral data were used to evaluate home pen aggression, including the number of agonistic interactions (AINX) and constituent aggressive actions, during a 3-h period (0800 to 1100 h) once per week and their change in relation to the baseline. Time-budget behaviors and postures were analyzed over eight 24-h periods (2 d/wk) using 10-min instantaneous scan sampling that focused on only the dominant and subordinate pigs in each pen. These 2 pigs were also subjected to blood collection once per week during the trial to determine concentrations of dopamine, norepinephrine, epinephrine, and serotonin (5-HT) using HPLC. Gilts performed more bites and total actions per AINX than barrows, and RAC-fed gilts increased bites and pursuits, whereas these behaviors decreased compared with baseline values in all other subgroups ($P < 0.05$). Gilts fed RAC increased the total number actions per AINX, whereas the occurrence of AINX decreased for all subgroups ($P < 0.01$). Overall, RAC-fed pigs were more behaviorally active, spending more time alert, bar biting, and sham chewing compared with CTL pigs ($P < 0.05$). The dominant RAC-fed pigs tended to have the greatest norepinephrine concentrations among the tested subgroups ($P = 0.08$). Dominant barrows had greater epinephrine concentrations than subordinate barrows ($P < 0.05$). The RAC-fed gilts tended to have lesser 5-HT concentrations than CTL gilts ($P = 0.08$), whereas concentrations were similar in barrows ($P > 0.10$). Greater activity and the increase in oral-related behaviors observed in RAC-fed pigs may be mediated by the increase in arousal caused by RAC. Intensified aggression in gilts, especially when fed RAC, may be linked to reduced central 5-HT and greater noradrenergic activity, and further research on brain neurotransmitters in gilts is needed.

Key words: aggression, amine, ractopamine, sex, social rank, swine

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INTRODUCTION

Ractopamine (**RAC**) is a widely used feed additive in swine production because it enhances production performance in finishing pigs (Gu et al., 1991a,b; See et al., 2004). Acting on β-adrenoreceptors, RAC augments lipolysis (Mills and Spurlock, 2003) and growth performance (See et al., 2004; Poletto et al., 2009). However, hyperactivity, overreactivity to transport, and difficulty in handling, in addition to elevated heart rate and increased peripheral catecholamine concentrations, have been reported in pigs fed RAC (Marchant-Forde et al., 2003). As a β-adrenoreceptor agonist, RAC mimics the effects of norepinephrine (**NE**) and epinephrine (**EPI**), enhancing arousal (Berridge, 2008); thus, RAC may have the potential to stimulate aggression.

The catecholaminergic system, which includes dopamine (**DA**), NE, and EPI, is among the first neu-

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rochemical systems to react during the fight-or-flight response (Haller et al., 1998). Increased behavioral arousal is particularly mediated by noradrenergic activation, which is also mimicked by RAC. Hence, RAC feeding may contribute to more frequent social interactions, thereby increasing the occurrence and severity of agonistic interactions. The serotonergic system, represented by serotonin (**5-HT**), is interconnected with the noradrenergic system (Clement et al., 1992) and is also linked to the regulation of aggression (Miczek and Fish, 2006). Thus, the effects of feeding RAC to finishing pigs on aggression, which has the potential to reduce productivity and well-being, warrants further investigation. The objectives of this study were to determine the effects of RAC on 1) the frequency of agonistic interactions and the constituent actions (bites, head knocks, and pursuits) displayed during each interaction in the home pen, 2) time-budget behavioral activity using scan sampling, and 3) alterations in peripheral concentrations of amines that are related to aggression. The effects of, and interactions with, sex and social rank were also investigated.

MATERIALS AND METHODS

The experimental procedures used in this study were approved by the Purdue University Animal Care and Use Committee and pigs were housed in accordance with FASS (1999) guidelines.

Animals and Housing

A total of 64 finishing pigs, including 32 barrows and 32 gilts [(US Duroc × Hampshire) × (US York × Landrace)], were selected from a population of 170 animals according to their initial BW. At mixing (d −14), experimental pigs were assigned to 1 of 4 groups of 4 pens, thus yielding a total of 16 pens on trial. Each group of pens was determined on the basis of similarity in initial BW of pigs and avoiding their parental relatedness. Within each group, there were 2 pens of only gilts and 2 pens of only barrows, which were assigned to 1 of the 2 dietary treatments. The average BW of the pigs across pens within a group was similar, but it varied slightly across groups. The average BW (±SEM) for groups were as follows: group 1 = 77.6 ± 0.6 kg, group 2 = 83.4 ± 0.1 kg, group 3 = 72.4 ± 0.2 kg, and group 4 = 77.5 ± 0.4 kg. Groups 1 and 2, or a total of 8 pens, went on trial simultaneously; groups 3 and 4 also went on trial simultaneously but 2 wk after groups 1 and 2. Each group was allocated to 1 of 2 rows of adjacent pens. As 2 groups went on trial at the same time, 1 group was assigned to 4 adjacent pens in the first row and the other group was assigned to 4 adjacent pens in the row on the opposite side, separated by a 1.5-m-wide aisle. Pigs belonging to all 4 groups were housed in the same barn. Thus, for a 2-wk overlapping period, all 16 pens were on trial at the same time. Pens within each group and within each row were organized so that they alternated sex and dietary treatment order to remove any potential location effects within the barn.

Each home pen was 1.8 × 3.0 m with the rear two-thirds of the floor being fully slatted concrete and the front one-third of the floor being plastic-coated expanded metal. A single nipple drinker was mounted in the middle of one of the pen sides, and a single-spaced feeder was situated at the front of each pen. The pigs were provided water and feed ad libitum. The room was ventilated naturally and the temperature was maintained at a minimum of 18.5°C because the trial was carried out during the winter (January to March); the external temperature never exceeded the room temperature during the trial period. Within group, pigs in each pen were assigned to the dietary treatments, which were either a control diet (**CTL**) or RAC feeding. Pigs on both dietary treatments were initially fed the same standard basal (CTL) diet for 2 wk (d −14 to 0). The basal diet was corn and soybean meal based (17.6% CP, 1.1% Lys) provided in meal form (as-fed basis). Thereafter, CTL pigs continued to receive the standard basal diet, whereas pigs assigned to the RAC dietary treatment had part of the starch fraction of the basal diet substituted with ractopamine hydrochloride (RAC, Paylean, Elanco Animal Health, Greenfield, IN). Ractopamine was delivered using a step-up feeding program in which RAC-fed pigs received 5 mg of RAC/kg of diet (as-fed basis) for 2 wk (phase I, d 0 to 14), and then 10 mg of RAC/kg of diet (as-fed basis) for the final 2 wk preceding slaughter (phase II, d 14 to 28). A detailed table with the composition of the experimental basal diet provided to the finishing pigs is available in Poletto et al. (2009).

Behavior Recording and Assignment of Social Rank

On pen assignment at mixing (d −14), pigs within each pen were individually marked for identification during behavioral observation. Behavior recording of all pigs in each pen began at the time of mixing (2 wk before the beginning of the dietary treatment) and continued for 36 h, using ceiling-mounted cameras (Panasonic WV-CD110AE, Matsushita Electric Industrial Co. Ltd., Osaka, Japan) attached to a digital video-recording system (IPD-DVR816, Inter-Pacific Inc., Northbrook, IL). No software was used to extract the behavioral data; an observer (the same person) watched all recorded videos. This behavioral information was used to investigate dominance hierarchy formation (i.e., the social rank of pigs within their new home pens).

The top dominant and the bottom subordinate pigs in each pen of 4 pigs were determined using continuous focal sampling of all pigs for the 36-h period, concentrating on the outcome of all agonistic social interactions (**AINX**) taking place within each pen. An AINX was defined as an event in which a pig directed offensive, forced behaviors against another pig in the group, during which the identification of initiators and receivers of the first attack were recorded. Offensive (i.e., bites, head knocks, pursuits, threats) and defensive behaviors (i.e., freeze, avoiding, flight) displayed and the duration of each AINX were recorded. A threat was defined as a pig, with or without its mouth open, displaying a vigorous lunging movement of its head toward another pig without making physical contact. A pig was deemed the loser of each AINX when it clearly showed any of the defensive behaviors mentioned that led to termination of the social interaction.

The following criteria were used to determine the social rank of pigs within each pen 1) dominance matrix: estimated by the number of occasions when a pig was supplanted vs. the number of occasions that that a pig supplanted others, using the outcomes of all pair-wise interactions (Martin and Bateson, 1993); and 2) level and success of the interaction: measured by counting the total frequency of AINX in which each pig was involved and the percentages of interactions that the pig won or lost, as evidenced by supplanting or resisting the displacement by the opponent (Bradshaw et al., 2000). Outcomes from both approaches, but more robustly, the level and success of the interaction, assisted in determining the top dominant (greatest scores, >0.66) and bottom subordinate (least scores, <0.33) pig in each pen. The remaining 2 pigs in the pen, with middle scores, were categorized as intermediate in social rank.

Home Pen Aggression

The behavior from all pigs in each pen was recorded continuously from 0800 to 1100 h (3-h period) once per week for 5 wk. The first recording was carried out on the week before beginning the dietary treatment, on d −2, and was used as baseline information for data analysis. Further recording was carried out on d 5 and 12 (phase I) and d 19 and 26 (phase II) of the trial. This behavioral information was also used to monitor the consistency of social rank of pigs in each pen.

To examine aggression in the home pen, the occurrence of AINX and constituent actions (i.e., bites, head knocks, and pursuits) displayed by the 2 pigs engaged in the AINX were recorded. Each AINX was defined as an event in which 1 or both pigs directed 1 or a sequence of offensive behaviors (bite, head knock, and pursuit), the AINX actions. The initiator of the first attack was also recorded. The total number of AINX, but also the number of each AINX constituent action (bites, head knocks, and pursuits) was recorded during the 3-h period. The total numbers of each action and their sum (bite + head knock + pursuit) were divided by the total number of AINX happening within that period, thus yielding the average number of actions per interaction; these averages were then used for statistical analysis. The percentage of changes in AINX occurrence, the number of each action, and their sum per AINX were also analyzed in relation to their respective baselines (d −2).

Behavioral Activity

Time-budget behavioral observations focused only on the top dominant and the bottom subordinate pigs in each pen (2 pigs per pen). Behavior was recorded continuously for a period of 24 h for 4 d during phase I (d 2, 5, 8, and 12) and an additional 4 d during phase II (d 15, 19, 22, and 26), yielding a total of eight 24-h behavior recordings. The video data were analyzed by an observer (the same person) using a 10-min instantaneous scan-sampling method, and the behavior and posture of the dominant and subordinate pigs in each pen were recorded at each scan. The description of inactive and active behaviors and postures are presented in the ethogram in Table 1. Inactive and all active behaviors were mutually exclusive at any time point of observation; at a specific time point, only 1 behavior (inactive or an active behavior) was assigned to each pig. The sum of inactive plus active behaviors added up to 100%. Additionally, the percentage of time spent in the specific body postures of lying, sitting, or standing was recorded. Postures were also mutually exclusive at any time point of observation; at a specific time point, only 1 posture was assigned to each pig. The sum of all postures added up to 100%. The only posture and behavior combinations that were not possible were lying + walking, sitting + walking, and lying + drinking.

For each pig, the proportions of time spent performing a behavior and in a specific posture within the hour in each day (within phases I and II) were divided by the total possible observations per hour (6 observations per pig; e.g., 2 out of 6 times a pig was inactive, thus 2/6 = 0.33), and multiplied by 100 to generate percentage values per hour (e.g., 33% of time spent inactive). The percentage of time spent performing a given behavior or in a specific posture per hour was then averaged across each day to yield a 24-h average that was used for statistical analysis. Thus, means of time-budget behaviors and postures are shown as the total percentage (%) of time spent performing the behaviors in a 24-h interval.

Blood Sample Collection and HLPC Assay

Blood samples were collected only from the top dominant and bottom subordinate pigs (2 pigs) in each pen once per week for 4 wk (phase I, d 4 and 11; phase II, d 18 and 25). The sampling procedure was initiated at the same time (0900 h) on all 4 d, and the collection order on each day was balanced by alternating sampling between dominant and subordinate pigs within each pen. Pigs were restrained with a wire metal nose snare and blood was collected into 10-mL BD Vacutainer K_3EDTA tubes (BD, Franklin Lakes, NJ) by jugular venipuncture. The tubes contained 0.117 mL of 15% K_3EDTA for a total of 17.55 mg. Aliquots of plasma (1 mL) were obtained by centrifugation at 760 × g for 15 min at 4°C, whereas whole blood samples were sepa-

Table 1. Ethogram used for evaluation of the time-budget behaviors and postures observed in the finishing pigs

Item	Description
Behavior	
Inactive	Physical immobility or without any activity
Alert	Ears positioned in a erect position of attention
Walking	Walking or running around the pen
Nosing or rooting	Manipulating with the snout any item in the pen (e.g., floor, bars)
Bar biting	Biting or chewing on the bars of the pen
Sham chewing	Chewing action performed without the presence of food in the mouth
Chain chewing	Biting or chewing on a metal chain hanging from the front gate
Nonaggressive interaction	Directing investigatory behavior toward another pig (e.g., sniffs, massage)
Drinking	Drinking water from the nipple drinker
Feeding	Head positioned inside of the feeder with oral movement
Posture	
Standing	Standing on all 4 legs
Lying	Lying down, either on the belly or on 1 side
Sitting	Dog-sitting with rump on the floor and shoulders raised up with front legs extended

rated into 0.5-mL aliquots. All samples were stored at −80°C until processing. Nonfocal (intermediate) pigs in each pen were similarly restrained and sham sampled by applying pressure on the jugular vein. The sham blood sampling was performed to handle and restrain all pigs in the pen equally in an attempt to prevent any potential effect of handling on the social structure of the group.

Peripheral concentrations of NE, EPI, and DA were measured using HPLC with a plasma catecholamine analysis kit (ESA Inc., Chelmsford, MA; Anton and Sayre, 1962). Samples were deproteinized and acidified with 100 µL of 4 *M* perchloric acid and then centrifuged at 13,000 × *g* for 10 min at 4°C. Acid supernatants were absorbed onto an alumina minicolumn, and 50 µL of dihydroxybenzylamine was added to be used as the internal standard. Columns were placed on a rocker for 10 min to allow catecholamines to bind to the alumina; HPLC minicolumns were then rinsed (INC Biomedicals Inc., Costa Mesa, CA) and eluted (ESA Inc.). Eluents were transferred into catecholamine tubes and placed on an autosampler set at 4°C. Each sample was double-injected into the reverse-phase columns using an ESA Coulochem II electrochemical detector (ESA Inc.). An ESA HR-80 column (80 mm in length) with a pore size of 120 Å was used in the study. The mobile phase (75 m*M* Na_2HPO_4, 1.7 m*M* 1-octanesulfonic acid, 25 µ*M* EDTA, 10% CH_3CN, and 100 µL/L of triethylamine, adjusted to pH 3.00 with phosphoric acid) was set at a flow rate of 0.9 mL/min. The concentrations (pg/mL) for NE, EPI, and DA were calculated from a reference curve constructed using the provided standards (Sigma-Aldrich, St. Louis, MO). The minimum detectable concentration for NE, EPI, and DA was 50 pg/mL. The intra- and interassay CV for NE, EPI, and DA analyses were 2.1 and 5.1%, respectively.

Peripheral Trp and 5-HT were also measured using HPLC. Whole blood samples were acidified using 100 µL of 4 *M* perchloric acid and freshly prepared ascorbic acid. After centrifugation at 13,000 × *g* for 5 min at 4°C, the acid supernatants were filtered and transferred to HPLC tubes and then placed into an autosampler set at 4°C. Each sample was double-injected onto a Linear Fluor LC-305 instrument (ESA Inc.) and column Platinum EPS C-18 column (Alltech Biotechnology, Nicholasville, KY) with a pore size 100 Å and 250 mm in length; the mobile phase flow rate was set at 1.2 mL/min. The concentrations (µg/mL) of Trp and 5-HT were calculated from a reference curve constructed using the provided standards (Sigma-Aldrich). The minimum concentration detectable for Trp and 5-HT was 1 µg/mL. The intra- and interassay CV for the Trp and 5-HT analyses were 1.5 and 2.2%, respectively.

Statistical Analysis

The experimental design consisted of 4 repeated groups of pens, with pens within each group housing pigs with similar initial BW. Groups 1 and 2 went on trial simultaneously but 2 wk before groups 3 and 4, which also went on trial simultaneously. Each group consisted of 4 pens, with each pen housing 4 pigs that were assigned to a dietary treatment (RAC or CTL), that belonged to the same sex (barrow or gilt), and that were balanced in an alternating fashion throughout the barn. For the analysis of home pen aggression data (total number of AINX; number of bites, head knocks, and pursuits; their sum per AINX; and their changes in relation to the baseline), which were collected based on AINX involving all 4 pigs within each pen, a 2 × 2 factorial arrangement of dietary treatment and sex was computed using pen (n = 16) as the experimental unit. All the variances were estimated, and model assumptions (e.g., normality) were assessed using the UNIVARIATE procedure (SAS Inst. Inc., Cary, NC). Data lacking normality were transformed using square root or $\log_{10}$, whichever was most appropriate. The MIXED procedure of SAS, with repeated measures of mixed models, was applied. The days of data collection pertaining to both phases I and II of the trial were nested within the corresponding phase [phase (day)] and were used as repeated measures. The analysis model included the terms of fixed effects for dietary treatment (CTL and RAC), sex (barrows and gilts), and their interaction (2-way); the main effect of phase (day); and the 2-way interactions of phase (day) × dietary treatment and phase (day) × sex. The random effects included the interaction of dietary treatment × sex nested within pen.

Social rank (dominant and subordinate status; 2 pigs per pen) was included as an additional model factor for the analyses of time-budget behavior and peripheral amine data. The inclusion of this factor generated a 2 × 2 × 2 factorial arrangement of dietary treatment, sex, and social rank with 4 repeated groups using pig (n = 32) as the experimental unit. There are limitations to considering pigs within pen as the experimental unit because these factors are not necessarily independent of each other. However, to test for the effects of social rank on variables measured within a pen, pig and not pen had to be used as the experimental unit. Treating individuals within the same group as experimental units may be deemed pseudoreplication (see the review by Hurlbert, 1984), but this can be handled statistically by nesting during analysis. To address the fact that each pen contained a dominant and subordinate pig that could have influenced each other behaviorally and physiologically, pen nested within group [group (pen)] was included in the model; thus, our analysis did not assume that samples from a given pen provided completely independent data. Several pig studies of a similar statistical design have been reported previously (e.g., McGlone et al., 1993; Morrow-Tesch et al., 1994; Sutherland et al., 2006; Poletto et al., 2009).

All the variances were estimated, and model assumptions (e.g., normality) were assessed using the UNIVARIATE procedure of SAS. Behavioral data lacking normality were square root transformed. The MIXED procedure of SAS, with repeated measures mixed models, was applied to analyze time-budget behaviors and the HPLC peripheral amine results, and phase (day) was applied as a repeated measure. The analysis model included the terms of fixed effects for dietary treatment, sex, social rank, and their interactions (3-way), the main effect of phase (day), and the 2-way interactions of phase (day) with dietary treatment, sex, and social rank. The random effects included the 3-way interaction of dietary treatment, sex, and social rank nested within pig and pen nested within group. For all analyses, main effects and all possible interactions were computed depending on the significance of the higher order interactions, and P-values were adjusted according to a Tukey post hoc test. All means, their respective SEM, and P-values are presented in tables and figures or are described in the text. Mean differences of $P < 0.05$ were considered statistically different, whereas mean differences of $P < 0.10$ were considered tendencies toward significance.

RESULTS

Home Pen Aggression Analysis

Bite Display per AINX. Gilts showed more bites per AINX than barrows (1.7 vs. 0.9 ± 0.2; $P < 0.05$) during the 3-h intervals observed each day. The interaction of treatment × sex was not significant for the number of bites per AINX ($P > 0.10$; Figure 1). However, the number of bites per AINX increased in relation to the baseline by almost 100% in RAC-fed gilts, whereas it decreased by an average of 35% for the other subgroups ($P < 0.001$; Table 2).

Head-Knock Display per AINX. Overall, CTL barrows showed the least number of head knocks per AINX when compared with CTL gilts, RAC-fed barrows, and RAC-fed gilts (treatment × sex, $P < 0.05$; Figure 1). In relation to the baseline, CTL barrows decreased the performance of head knocks per AINX, whereas CTL gilts and RAC barrows increased head knocks ($P < 0.05$); RAC-fed gilts also decreased the number of head knocks per AINX in relation to the baseline, but this change was slight (treatment × sex, $P < 0.01$; Table 2).

Pursuit Display per AINX. Gilts tended to perform more pursuits per AINX on d 5, 12 (phase I), and 26 (phase II) but fewer pursuits on d 19 compared with barrows at the same time points [sex × phase (day), $P = 0.08$]. There was no evidence of significant main effects or a treatment × sex interaction for the number of pursuits per AINX ($P > 0.10$; Figure 1), but the number of pursuits in relation to the baseline increased by more than 300% in the RAC-fed gilts, whereas it decreased for the other subgroups (treatment × sex, $P < 0.001$; Table 2).

Total Actions Display per AINX. Gilts had more total actions per AINX than barrows (2.5 vs. 1.5 ± 0.2, $P < 0.01$). Furthermore, in relation to the baseline, RAC-fed gilts had an increase in the number of total actions (bites + head knocks + pursuits) performed per AINX compared with decreases in all the other subgroups (treatment × sex, $P < 0.01$; Table 2).

Total Number of AINX. When analyzing the total number of AINX in relation to the baseline, the treatment × sex interaction tended to be significant ($P = 0.07$), with no statistical trend being detected for pair-wise comparisons ($P > 0.10$). All 4 subgroups decreased the number of AINX in which they engaged by an average of 60%; CTL gilts showed the smallest reduction in the total number of AINX, whereas CTL barrows showed the greatest reduction ($P > 0.10$; Table 2).

Active Behaviors and Postures

Overall, RAC-fed pigs were more behaviorally active (i.e., showed a greater state of arousal) compared with CTL pigs ($P < 0.05$; Table 3), and this difference became evident toward to the end of phase I (d 12) and

Table 2. Percentage of changes in constituent actions displayed during agonistic interactions (AINX) and the total number of AINX in relation to the respective baseline counts, according to the dietary treatment[1] and sex of the finishing pigs

	CTL		RAC	
Action,[2] %	Barrow	Gilt	Barrow	Gilt
Bites/AINX	↓47.3^{b}	↓35.0^{b}	↓20.5^{b}	↑95.7^{a}
Head knocks/AINX	↓35.4^{a}	↑21.3^{b}	↑14.1^{b}	↓4.1^{ab}
Pursuits/AINX	↓0.5^{b}	↓76.4^{b}	↓62.5^{b}	↑334.6^{a}
Total actions/AINX	↓44.0^{b}	↓24.4^{b}	↓10.0^{b}	↑54.9^{a}
Total AINX	↓66.4^{a}	↓50.1^{a}	↓61.0^{a}	↓64.5^{a}

[a,b]Within a row and between dietary treatments and sexes, means without a common superscript letter differ ($P < 0.05$); treatment × sex, $P < 0.01$.

[1]CTL = pigs fed the control diet; RAC = pigs fed the ractopamine-added (Elanco Animal Health, Greenfield, IN) diet.

[2]In relation to the respective baseline averages observed 2 d before the beginning of the feeding trial (d −2); arrows pointing up (↑) represent a percentage increase in actions displayed, whereas arrows pointing down (↓) represent a percentage decrease.

through most of phase II [treatment × phase (day), $P < 0.05$; Figure 2]. No evidence of differences was observed during the first week of RAC feeding ($P > 0.10$). Furthermore, an increasing pattern of alertness was observed in RAC-fed pigs throughout the experiment, with the pigs becoming different statistically from the CTL pigs during phase II [treatment × phase (day), $P < 0.01$; Figure 3]. The percentage of time spent walking was not affected by RAC feeding, sex, or social rank (Table 3).

The differences in activity levels observed in RAC-fed pigs were mostly associated with the performance of certain oral-nasal behaviors. Pigs fed RAC spent more time sham chewing, bar biting, and feeding than the CTL pigs ($P < 0.05$; Table 3). Dominant RAC-fed pigs tended to spend more time bar biting compared with dominant CTL pigs (0.6 vs. 0.3 ± 0.1%, $P = 0.07$), but neither differed from subordinate CTL and RAC-fed pigs (0.4 and 0.4 ± 0.1%, respectively; treatment × social rank, $P = 0.08$). Gilts spent more time bar biting and nosing or rooting than barrows ($P < 0.05$), whereas barrows visited the drinker more often than gilts ($P < 0.05$; Table 3). Time spent performing nosing or rooting also increased during the middle of both phase I (d 5 and 8) and phase II (d 19 and 22) for all pigs [phase (day), $P < 0.001$]. Dominant pigs, when compared with subordinate pigs, tended to spend more time manipulating the metal chain located on the front

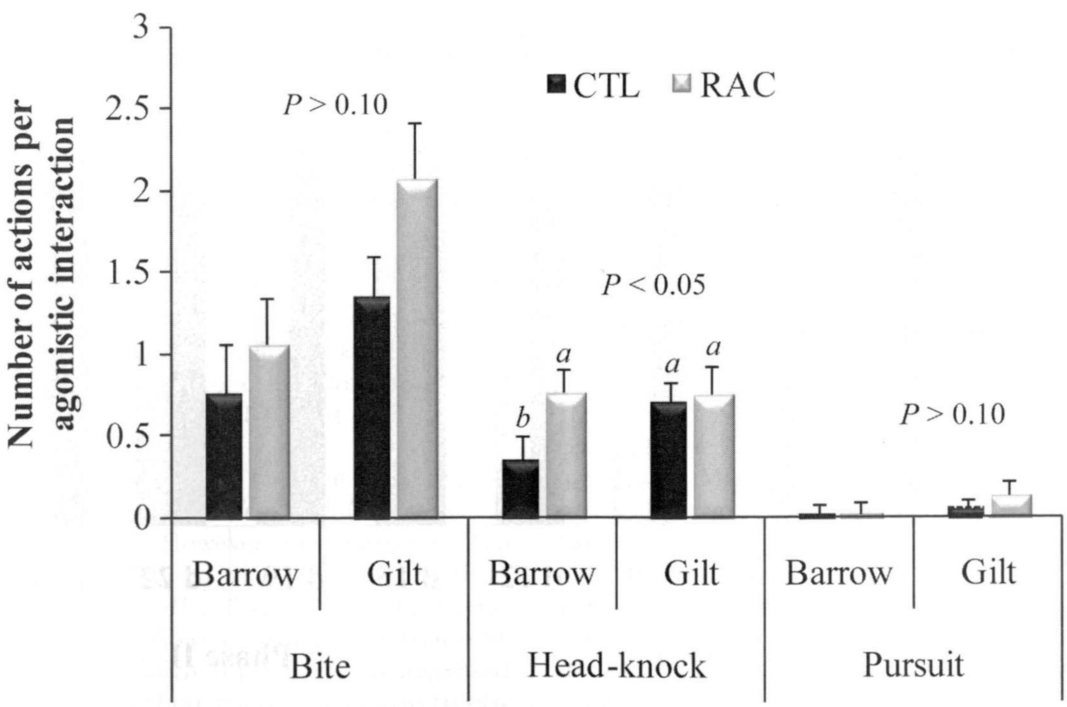

Figure 1. Overall average number of bites, head knocks, and pursuits per agonistic interaction displayed by control (CTL) and ractopamine-fed (RAC, Elanco Animal Health, Greenfield, IN) barrows and gilts during a 3-h continuous observation period (0800 to 1100 h) per week for a 4-wk feeding trial. Error bars represent the SEM. $^{a,b}P < 0.05$.

Table 3. Mean percentages of time spent in various behaviors and postures, shown as averages for a 24-h interval, by finishing pigs according to ractopamine dietary treatment, sex, and social rank

	Treatment[1]		Sex		Social rank		
Item	CTL	RAC	Barrow	Gilt	Dominant	Subordinate	Pooled SEM
Behavior, %							
Inactive	77.8[a]	73.9[b]	75.9	75.8	76.5	75.2	1.8
Alert	2.2[a]	4.4[b]	3.5	3.3	3.3	3.6	0.6
Walking	0.7	0.8	0.7	0.7	0.7	0.7	0.1
Nosing or rooting	7.0	6.2	5.8[a]	7.4[b]	6.8	6.4	0.4
Chain chewing	1.1	1.0	0.9	1.2	1.3[x]	0.8[y]	0.2
Bar biting	0.3[a]	0.5[b]	0.3[a]	0.5[b]	0.4	0.4	0.0
Sham chewing	2.0[a]	2.7[b]	2.5	2.2	2.5	2.4	0.2
Nonaggressive interactions	2.1	2.7	2.6	2.2	1.9[x]	2.9[y]	0.4
Feeding	5.3[a]	6.2[b]	6.0	5.4	5.5	6.0	0.3
Drinking	0.8	0.9	1.0[a]	0.7[b]	0.7[x]	0.9[y]	0.1
Posture, %							
Standing	15.1	16.0	14.6	16.5	14.8	16.3	0.9
Sitting	0.9[a]	2.4[b]	1.4	1.9	1.7	1.6	0.4
Lying	84.0[x]	81.7[y]	84.0[x]	81.6[y]	83.6	82.1	0.9

[a,b]Within a row and between dietary treatments, sexes, and social ranks, means without a common superscript letter differ ($P < 0.05$).
[x,y]Within a row and between dietary treatments, sexes, and social ranks, means without a common superscript letter differ ($P < 0.10$).
[1]CTL = pigs fed the control diet, RAC = pigs fed the ractopamine-added (Elanco Animal Health, Greenfield, IN) diet.

gate of the pen and less time at the drinker ($P = 0.08$; Table 3). When evaluating nonaggressive social interactions, subordinate pigs tended to initiate more physical exploration toward another pen mate compared with dominant pigs ($P = 0.09$; Table 3). The effect of phase (day) was significant for nonaggressive interactions ($P < 0.05$), which increased during the middle of both phase I (d 5 and 8) and phase II (d 19), following a pattern similar to that observed for the performance of nosing or rooting behavior. Regarding the posture analysis, RAC-fed pigs spent more time sitting ($P < 0.05$) and tended to spend less time lying than the CTL pigs ($P = 0.07$; Table 3). Gilts also tended to spend less time lying ($P = 0.07$) than barrows; subordinate gilts tended to spend the most time sitting overall (2.3 ± 0.6) compared with dominant barrows, dominant gilts, and subordinate barrows (1.8, 1.4, 1.0 ± 0.6, respectively; sex × social rank, $P = 0.08$). Across phases, dominant pigs spent significantly more time sitting on d 22 (2.8 ± 0.4) than on d 12, 15, and 19 (0.9, 1.2, 0.9 ±

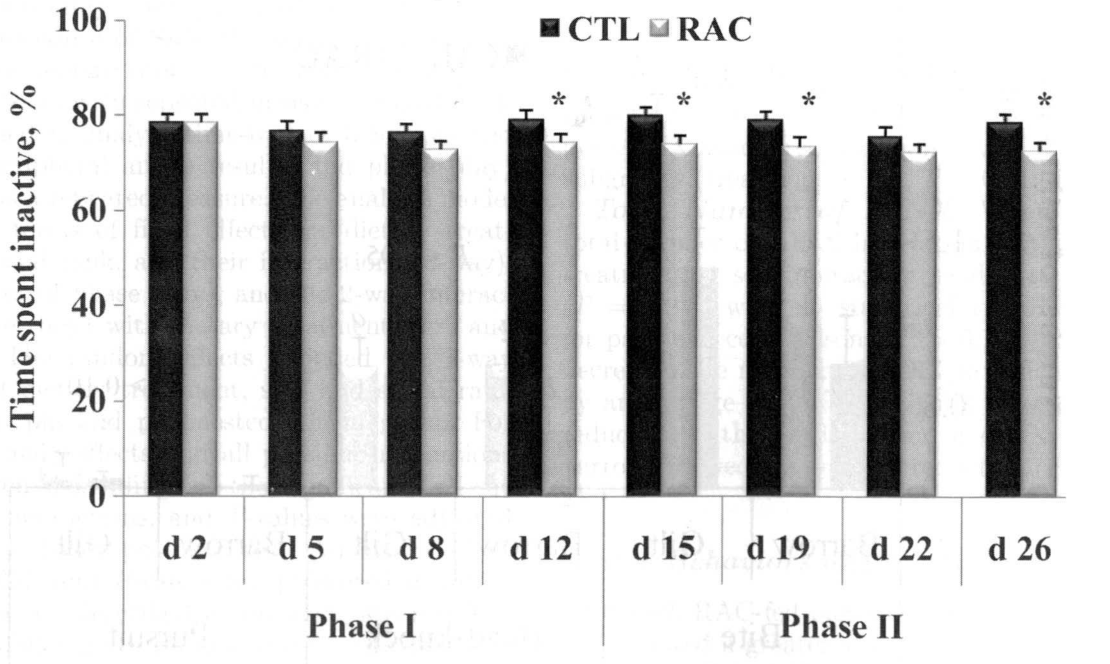

Figure 2. Percentage of time spent inactive by finishing pigs fed either the control (CTL) or the ractopamine-added (RAC, Elanco Animal Health, Greenfield, IN) diet. Phase I represents the 2 wk when pigs received RAC at 5 mg/kg of diet (as-fed basis), and phase II represents the final 2 wk when pigs were fed RAC at 10 mg/kg of diet (as-fed basis). Error bars represent the SEM. *$P < 0.05$.

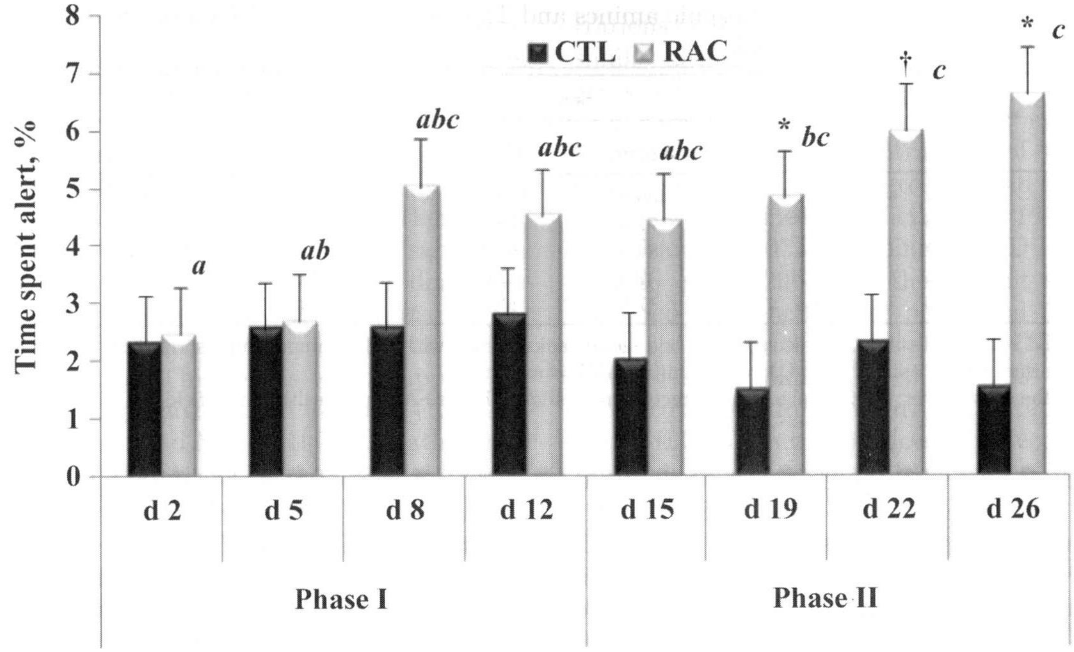

Figure 3. Percentage of time spent alert by finishing pigs fed either the control (CTL) or the ractopamine-added (RAC, Elanco Animal Health, Greenfield, IN) diets. Phase I represents the 2 wk when pigs received RAC at 5 mg/kg of diet (as-fed basis), and phase II represents the final 2 wk when pigs were fed RAC at 10 mg/kg of diet (as-fed basis). Error bars represent the SEM. †$P < 0.10$; *$P < 0.05$ between dietary treatments. [a–c]$P < 0.05$ over time within RAC-fed pigs.

0.4, respectively, $P < 0.05$), but they were not different from subordinate pigs at the same time points [social rank × phase (day), $P < 0.05$].

Blood Profile for Amines and Tryptophan

Dominant RAC-fed pigs tended to have the greatest plasma NE concentrations, whereas dominant CTL pigs had the least plasma NE concentrations (465.6 vs. 331.2 ± 64.6 pg/mL, respectively, $P = 0.08$); subordinate CTL and RAC-fed pigs had intermediate NE concentrations (382.9 and 380.6 ± 64.6 pg/mL, respectively; treatment × social rank, $P = 0.08$). In addition, dominant barrows had greater plasma EPI concentrations than subordinate barrows (94.5 vs. 61.1 ± 9.8 pg/mL, respectively, $P < 0.05$); however, dominant and subordinate gilts had similar EPI concentrations (61.2 and 70.7 ± 9.8 pg/mL, respectively; sex × social rank, $P < 0.05$). The DA concentrations increased consistently for all pigs during the trial [phase (day), $P < 0.001$], reaching the greatest concentration on d 18 (d 4 = 43.5, d 11 = 97.6, d 18 = 135.5, and d 25 = 113.5 ± 16.2 pg/mL).

Ractopamine-fed gilts tended to have lesser blood 5-HT concentrations than CTL-fed gilts (treatment × sex, $P = 0.08$; Figure 4). However, no changes in Trp concentrations were associated with RAC feeding (Figure 4), sex, or social rank ($P > 0.10$). Blood concentrations of Trp and 5-HT followed a similar pattern over the days within phases; both concentrations increased up to d 11 (phase I), and these concentrations were sustained until the end of the experiment. Concentrations of 5-HT were d 4 = 2.0, d 11 = 2.4, d 18 = 2.4, and d 25 = 2.5 ± 0.1 μg/mL, and concentrations of Trp were d 4 = 9.8, d 11 = 11.8, d 18 = 11.7, and d 25 = 12.5 ± 0.4 μg/mL [phase (day), $P < 0.001$]. The main effect means for the biogenic amines and Trp are presented in Table 4.

DISCUSSION

This study shows that provision of RAC as a feed additive to finishing pigs increases behavioral activity, including alertness and performance of oral-nasal behaviors. Furthermore, RAC increases the frequency of aggressive behaviors and the severity of the AINX in gilts, as evidenced by the greater number of offensive behaviors, particularly bites. This outcome may be linked to the availability and action of neurotransmitters such as NE and 5-HT, which are essential for controlling aggression in the central nervous system (e.g., Haller et al., 1998; Miczek and Fish, 2006). Peripherally, there was hardly any alteration in amine concentrations in gilts, but dominant RAC-fed pigs had the greatest NE plasma concentration; social rank also played a role in EPI concentrations. In agreement with a study by Marchant-Forde et al. (2003), feeding RAC made the pigs hyperactive; this effect became visible from the second week of feeding at 5 mg of RAC/kg of diet and was sustained until the end of the trial, during which RAC was provided at 10 mg/kg of diet. Although RAC-fed pigs visited the feeder more often, no differences in ADFI between treatments were observed, as reported previously by Poletto et al. (2009), who studied the same experimental pigs used in the current study. Instead, a greater ADG was seen in the RAC-fed

Table 4. Peripheral concentrations of biogenic amines and Trp measured in the blood of finishing pigs, in relation to dietary treatment, sex, and social rank

	Treatment[2]		Sex		Social rank		
Analyte[1]	CTL	RAC	Barrow	Gilt	Dominant	Subordinate	Pooled SEM
NE, pg/mL	357.1	423.1	400.9	379.3	398.4	381.8	57.9
EPI, pg/mL	71.5	78.7	77.5	73.0	79.1	71.1	8.2
DA, pg/mL	86.1	85.4	84.0	87.4	84.0	87.5	11.4
5-HT, μg/mL	2.1	2.2	2.4^{x}	2.0^{y}	2.3	2.1	0.1
Trp, μg/mL	11.6	11.3	11.8	11.1	11.5	11.3	0.5

[x,y]Within a row and between dietary treatments, sexes, and social ranks, means without a common superscript letter differ ($P = 0.09$).
[1]NE = norepinephrine; EPI = epinephrine; DA = dopamine; 5-HT = serotonin.
[2]CTL = pigs fed the control diet; RAC = pigs fed the ractopamine-added (Elanco Animal Health, Greenfield, IN) diet.

(1.044 kg/d) compared with the CTL pigs (0.898 kg/d; Poletto et al., 2009). The greatest effect of RAC on the growth performance of pigs has been shown to occur in the first 14 d of feeding, and by sustaining the feeding dose of the compound, there is a gradual decrease in response caused by receptor desensitization or downregulation (Williams et al., 1994). Behaviorally, doubling RAC dietary concentration in the food provided to the pigs 2 wk after the beginning of the trial led to greater behavioral activity. This outcome is likely the result of sustained activation of β-adrenoreceptors mediated by the compound.

Greater activity was not correlated with more time spent walking because this was similar between the RAC-fed and CTL pigs. Instead, hyperactivity of RAC-fed pigs was associated with enhanced alertness, which increased progressively during the step-up RAC feeding program of 5 up to 10 mg/kg of diet, and the performance of oral-nasal behaviors such as sham chewing and bar biting. The increase in alert behavior was anticipated because RAC acts as a β-adrenoreceptor agonist and mimics the effects of the NE and EPI, neurotransmitters responsible for the neuroendocrine activation of the sympathetic system, leading to increases in arousal (Berridge, 2008). However, the mechanism for the increase in oral-nasal behaviors in the RAC-fed pigs is unknown. Other β-adrenoreceptor agonists, such as β_3 agonists, inhibit gastric acid secretion, minimizing the occurrence of stomach ulcers (Daly, 1984; Sevak et al., 2002). Thus, it may be speculated that the increase in performance of oral manipulatory behaviors increases saliva production, which may buffer stomach pH (Marchant-Forde and Pajor, 2003). However, because there is a potential for RAC to induce greater responsiveness to stress, as shown by Marchant-Forde et al. (2003), its role in potentially triggering stereotypic behaviors, such as sham chewing and bar biting, cannot be ignored. Difficulty of handling, elevated heart rate, and increased peripheral catecholamine concentrations reported in the RAC-fed pigs are indicators of over-responsiveness to stress (Marchant-Forde et al., 2003). Another β-adrenoreceptor agonist, isoproterenol, also physiologically modulates the β_2-adrenergic-positive feedback loop, which increases release of endogenous NE and potentiates behavioral reactivity to stress (Krantz et al., 1987).

On the evaluation of agonistic interactions in the home pen, carried out from continuous behavioral observations, gilts delivered twice as many bites per AINX than barrows. When fed RAC, gilts engaged in fewer agonistic interactions in relation to the baseline, but there was a substantial increase in the number of bites and pursuits per AINX in relation to the baseline. This behavioral pattern shows that AINX became less frequent but more severe because bites and pursuits can be considered retaliatory behaviors during fights. The

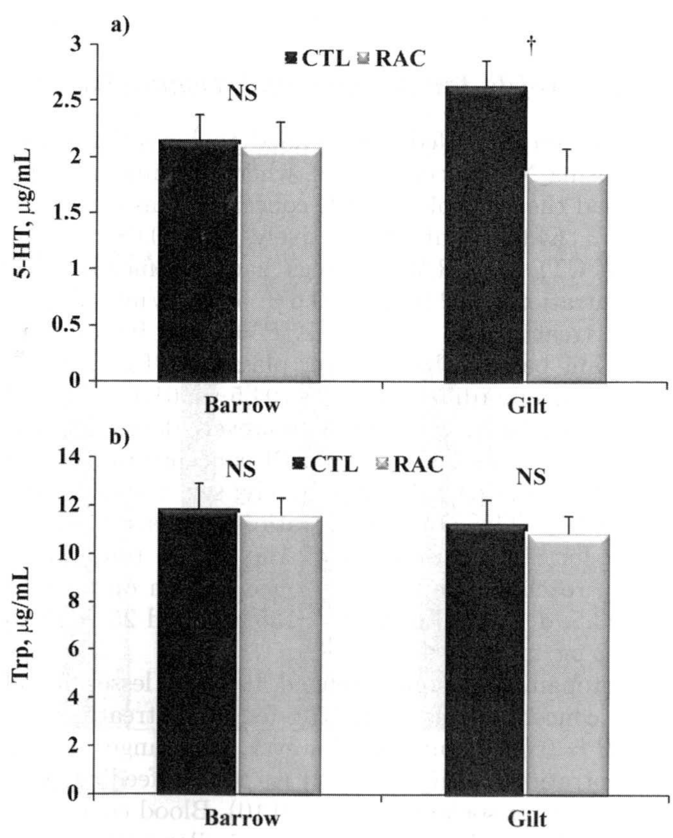

Figure 4. Peripheral blood concentrations of serotonin (5-HT; panel a) and Trp (panel b) in finishing barrows and gilts fed either the control (CTL) or ractopamine-added (RAC, Elanco Animal Health, Greenfield, IN) diets during a 4-wk feeding trial. Error bars represent the SEM. †$P = 0.08$; NS = not significant.

lack of evidence for the effect of feeding phase on agonistic behavior performance indicates that there was a homogeneous effect of RAC throughout the 4 wk of the experiment. Because pen averages were evaluated for home pen aggression, the effect of social rank status on home pen agonistic interactions could not be determined in the current study. Nevertheless, another study by Poletto et al. (2008), carried out with the same pigs used in the present study, reported that the dominant and subordinate RAC-fed gilts as well as dominant CTL gilts displayed the greatest frequency of attacks during a resident-intruder test. This test is widely applied to measure levels of aggressiveness in rodents (Kemble, 1993) and pigs (Erhard and Mendl, 1997; D'Eath, 2002; D'Eath and Pickup, 2002). Social rank of pigs played a role in passive-exploratory social behaviors, as shown by subordinate pigs spending approximately twice the amount of time directing nonaggressive behaviors such as nosing or sniffing toward other pigs in the pen. This leads us to speculate that directing more nonoffensive physical contact toward a pen mate may be disruptive and may be one factor that triggers more frequent agonistic interactions in the home pen.

A positive relationship exists between noradrenergic activity and fighting or biting behavior in various rodents and monkeys (Haller et al., 1998). This action is mediated by activation of the sympathetic nervous system, which mediates arousal and can increase the intensity and characteristics of anger-related emotional behaviors (Krantz et al., 1987). Although it is not known whether RAC crosses the blood-brain barrier, it is thought that catecholamines do not cross into the brain (Kostrzewa, 2007). However, the transport of aromatic AA precursors for the synthesis of the biogenic amines (i.e., as Trp is for 5-HT) across the blood-brain barrier is upregulated by β_2-adrenoceptors through administration of β-adrenoreceptor agonists (Edwards et al., 1989; Takao et al., 1992). According to Marchant-Forde et al. (2003), the concentrations of catecholamines at the end of a RAC feeding trial were greater in pigs fed the compound at a constant dose of 10 mg/kg of diet for 4 wk. However, in the current study a similar effect was not observed. It was expected that increasing the RAC dose after the initial 2 wk would minimize any potential desensitization of the adrenoreceptor (Williams et al., 1994) and that this would be reflected in the peripheral concentrations of the catecholamines. Instead, only a mild increase in NE concentration was detected in the dominant RAC-fed pigs, and no effect of feeding phase was detected. Sex and social rank appeared to influence the peripheral catecholamine profile of pigs more strongly than RAC feeding because dominant barrows had greater plasmatic concentrations of EPI than subordinate ones. Increased EPI and decreased NE concentrations characterize the activation of the sympathetic nervous system and are correlated with the maintenance of the social rank status and social competence (Dillon et al., 1992). However, in another study carried out in pigs, plasma NE and EPI concentrations of dominant and subordinate individuals were similar although they were linearly correlated with aggressive behaviors (Fernandez et al., 1994).

A deficiency in the serotonergic system, generally portrayed by low concentrations of 5-HT, has been widely associated with aggression and impulsive behaviors in several species (Miczek and Fish, 2006; Nelson and Trainor, 2007), including pigs (D'Eath et al., 2005; Poletto et al., 2008). In the current study, gilts that were fed RAC had lesser peripheral 5-HT concentrations than gilts that received the CTL diet, whereas there was no indication that RAC feeding affected 5-HT concentrations in barrows. Although the peripheral 5-HT concentration does not necessarily replicate the brain concentrations of the amine, these results are consistent with the fact that RAC-fed gilts displayed more aggressive behaviors during AINX in their home pens. The β_2-adrenoceptor agonist clenbuterol, when provided to rats at 5 mg/kg, significantly reduced plasma Tyr, the DA precursor, and raised brain Trp concentrations, the precursor for 5-HT (Edwards et al., 1989). In the current study, the whole blood concentration for Trp increased until d 11 of phase I, and Trp concentrations were sustained throughout the trial and were not affected by the dietary treatment, sex, or social rank of the pigs used for this study. The parallel increase in the concentrations of Trp and 5-HT, but also the increase in DA concentration until d 18, may be correlated with an increase in overall feed intake by the experimental pigs (Poletto et al., 2009). According to the NRC (1998), the minimum Trp inclusion rate required for pigs at the finishing phase is 0.11% of the total diet. However, RAC feeding requires all dietary AA to be increased by 50% in the diet because of an increase in muscle accretion caused by the effects of RAC (Schinckel et al., 2003). Thus, Trp was provided in the diet for both CTL and RAC groups at a 0.20% inclusion rate to ensure it was not limiting the RAC response (Poletto et al., 2009).

In summary, this study showed that RAC feeding caused finishing pigs to become more behaviorally active, which included more time spent alert, bar biting, and sham chewing, and more time spent sitting than lying. Ractopamine feeding of gilts, but not barrows, increased aggressive actions per AINX, such as bites and pursuits, whereas the number of AINX decreased in relation to pretrial behavior, suggesting an augmentation in the severity of interactions. Dominant social status was associated with greater plasmatic NE concentrations in the RAC-fed pigs and greater EPI concentrations in barrows. Ractopamine feeding decreased peripheral blood 5-HT concentrations in gilts, whereas Trp concentrations were unaltered. The increase in behavioral activity along with a greater sympathetic activity and less 5-HT availability may be linked to greater aggressiveness in RAC-fed pigs, especially in gilts. Future research should focus on elucidating the underlying mechanisms (i.e., at the central nervous system level) of intensified aggression in gilts and its association with RAC feeding, thus potentially minimizing production costs and impairment of pig well-being.

LITERATURE CITED

Anton, A. H., and D. F. Sayre. 1962. A study of the factors affecting the aluminum oxidetrihydroxyindole procedure for the analysis of catecholamines. J. Pharmacol. Exp. Ther. 138:360–375.

Berridge, C. W. 2008. Noradrenergic modulation of arousal. Brain Res. Rev. 58:1–17.

Bradshaw, R. H., J. Skyrme, E. E. Brenninkmeijer, and D. M. Broom. 2000. Consistency of measurement of social status in dry-sows group-housed in indoor and outdoor systems. Anim. Welf. 9:75–79.

Clement, H.-W., D. Gemsa, and W. Weseman. 1992. The effect of adrenergic drugs on serotonin metabolism in the nucleus raphe dorsalis of the rat, studies by in vitro voltammetry. Eur. J. Pharmacol. 217:43–48.

Daly, M. J. 1984. The classification of adrenoceptors and their effects on gastric acid secretion. Scand. J. Gastroenterol. Suppl. 89:3–9.

D'Eath, R. B. 2002. Individual aggressiveness measured in a resident-intruder test predicts the persistence of aggressive behaviour and weight gain of young pigs after mixing. Appl. Anim. Behav. Sci. 77:267–283.

D'Eath, R. B., E. Ormandy, A. B. Lawrence, B. E. H. Sumner, and S. L. Meddle. 2005. Resident-intruder trait aggression is associated with differences in lysine vasopressin and serotonin receptor 1A (5-HT1A) mRNA expression in the brain of prepubertal female domestic pigs (*Sus scrofa*). J. Neuroendocrinol. 17:679–686.

D'Eath, R. B., and H. E. Pickup. 2002. Behavior of young pigs in a resident-intruder test designed to measure aggressiveness. Aggress. Behav. 28:401–415.

Dillon, J. E., M. J. Raleigh, M. T. McGuire, D. Bergin-Pollack, and A. Yuwiler. 1992. Plasma catecholamines and social behavior in male vervet monkeys (*Cercopithecus aethiops sabaeus*). Physiol. Behav. 51:973–977.

Edwards, D. J., D. A. Sorisio, and S. Knopf. 1989. Effects of the β_2-adrenoceptor agonist clenbuterol on tyrosine and tryptophan in plasma and brain of the rat. Biochem. Pharmacol. 38:2957–2965.

Erhard, H. W., and M. Mendl. 1997. Measuring aggressiveness in growing pigs in a resident-intruder situation. Appl. Anim. Behav. Sci. 54:123–136.

FASS. 1999. Guide for the Care and Use of Agricultural Animals in Agricultural Research and Teaching. 1st rev. ed. Fed. Anim. Sci. Soc., Champaign, IL.

Fernandez, X., M. C. Meunier-Salaün, and P. Mormede. 1994. Agonistic behavior, plasma stress hormones, and metabolites in response to dyadic encounters in domestic pigs: Interrelationships and effect of dominance status. Physiol. Behav. 56:841–847.

Gu, Y., A. P. Schinckel, J. C. Forrest, C. H. Kuei, and L. E. Watkins. 1991a. Effects of ractopamine, genotype, and growth phase on finishing performance and carcass value in swine. I. Growth performance and carcass merit. J. Anim. Sci. 69:2685–2693.

Gu, Y., A. P. Schinckel, J. C. Forrest, C. H. Kuei, and L. E. Watkins. 1991b. Effects of ractopamine, genotype, and growth phase on finishing performance and carcass value in swine: II. Estimation of lean growth rate and lean feed efficiency. J. Anim. Sci. 69:2694–2702.

Haller, J., G. B. Makara, and M. R. Kruk. 1998. Catecholaminergic involvement in the control of aggression: Hormones, the peripheral sympathetic, and central noradrenergic systems. Neurosci. Biobehav. Rev. 22:85–97.

Hurlbert, S. H. 1984. Pseudoreplication and the design of ecological field experiments. Ecol. Monogr. 54:187–211.

Kemble, E. D. 1993. Resident-intruder paradigms for the study of rodent aggression. Pages 138–150 in Methods in Neurosciences: Paradigms for the Study of Behavior. Vol. 14. P. M. Conn, ed. Academic Press, New York, NY.

Kostrzewa, R. M. 2007. The blood-brain-barrier for catecholamines—Revisited. Neurotox. Res. 11:261–272.

Krantz, D. S., R. J. Contrada, P. J. LaRiccia, J. R. Anderson, L. A. Durel, T. M. Dembroski, and T. Weiss. 1987. Effects of beta-adrenergic stimulation and blockade on cardiovascular reactivity, affect, and type A behavior. Psychosom. Med. 2:146–158.

Marchant-Forde, J. N., D. C. Lay Jr., E. A. Pajor, B. T. Richert, and A. P. Schinckel. 2003. The effects of ractopamine on the behavior and physiology of finishing pigs. J. Anim. Sci. 81:416–422.

Marchant-Forde, J. N., and E. A. Pajor. 2003. The effects of dietary sodium bicarbonate on abnormal behavior and heart rate in sows. J. Anim. Sci. 81(Suppl 1):158. (Abstr.)

Martin, P., and P. P. G. Bateson. 1993. Measuring Behavior. Cambridge University Press, Cambridge, UK.

McGlone, J. J., J. L. Salak, E. A. Lumpkin, R. I. Nicholson, M. Gibson, and R. L. Norman. 1993. Shipping stress and social status effects on pig performance, plasma cortisol, natural killer cell activity and leukocyte numbers. J. Anim. Sci. 71:888–896.

Miczek, K. A., and E. W. Fish. 2006. Biology of Aggression. R. J. Nelson, ed. Oxford University Press New York, NY.

Mills, S. E., and M. E. Spurlock. 2003. β-Adrenergic subtypes that mediate ractopamine lipolysis. J. Anim. Sci. 81:662–668.

Morrow-Tesch, J. L., J. J. McGlone, and J. L. Salak-Johnson. 1994. Heat and social stress effects on pig immune measures. J. Anim. Sci. 72:2599–2609.

Nelson, R. J., and B. C. Trainor. 2007. Neural mechanisms of aggression. Nat. Rev. Neurosci. 8:536–546.

NRC. 1998. Nutrient Requirements of Swine. 10th ed. Natl. Acad. Press, Washington, DC.

Poletto, R., B. T. Richert, H. W. Cheng, R. L. Meisel, and J. N. Marchant-Forde. 2008. Effects of ractopamine feeding, gender and social rank on aggressiveness and monoamine concentrations in different brain areas of finishing pigs. Page 83 in Proc. 42nd Congr. Int. Soc. Appl. Ethol., Dublin, Ireland. L. Boyle, N. O'Connell, and A. Hanlon, ed. Wageningen Acad. Press, Wageningen, the Netherlands.

Poletto, R., M. H. Rostagno, B. T. Richert, and J. N. Marchant-Forde. 2009. Effects of ractopamine feeding, gender and social rank on growth performance, hoof lesions and *Enterobacteriaceae* shedding in finishing pigs. J. Anim. Sci. 87:304–313.

Schinckel, A. P., N. Li, B. T. Richert, P. V. Preckel, and M. E. Einstein. 2003. Development of a model to describe the compositional growth and dietary lysine requirements of pigs fed ractopamine. J. Anim. Sci. 81:1106–1119.

See, M. T., T. A. Armstrong, and W. C. Weldon. 2004. Effect of a ractopamine feeding program on growth performance and carcass composition in finishing pigs. J. Anim. Sci. 82:2474–2480.

Sevak, R., A. Paul, S. Goswami, and D. Santani. 2002. Gastroprotective effect of β_3 adrenoreceptor agonists ZD 7114 and CGP 12177A in rats. Pharmacol. Res. 46:351–356.

Sutherland, M. A., S. R. Niekamp, S. L. Rodriguez-Zas, and J. L. Salak-Johnson. 2006. Impacts of chronic stress and social status on various physiological and performance measures in pigs of different breeds. J. Anim. Sci. 84:588–596.

Takao, Y., Y. Kamisaki, and T. Itoh. 1992. Beta-adrenergic regulation of amine precursor amino acid transport across the blood-brain barrier. Eur. J. Pharmacol. 215:245–251.

Williams, N. H., T. R. Cline, A. P. Schinckel, and D. J. Jones. 1994. The impact of ractopamine, energy intake, and dietary fat on finisher pig growth performance and carcass merit. J. Anim. Sci. 72:3152–3162.